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New Spectrophotometric Assay for the Determination of Selected Drugs In Pure and Dosage Forms

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Abstract:

The present study focuses on the development and validation of simple, sensitive, accurate, and reproducible analytical methods for the quantitative determination of Meropenem in bulk drug and pharmaceutical dosage forms. Visible spectrophotometric methods were developed based on various chromogenic reactions involving reagents such as DDQ, p-chloranilic acid, 4-aminophenazone, isoniazid, anthranilic acid, 2-chlorophenyl hydrazine, 2,4-dinitrophenylhydrazine, bromothymol blue, bromocresol purple, and MBTH in the presence of Fe(III). These reagents form stable colored complexes with Meropenem, which were measured at their respective absorption maxima. Optimization of experimental conditions including reagent concentration, pH, temperature, reaction time, and solvent system was carried out to achieve maximum color intensity and stability. The developed methods obeyed Beer–Lambert's law within specific concentration ranges, demonstrating good linearity with high correlation coefficients. Analytical parameters such as accuracy, precision, sensitivity, and reproducibility were evaluated in accordance with standard validation guidelines. The proposed methods were successfully applied to the analysis of Meropenem in pharmaceutical formulations, and the results were found to be in good agreement with reference methods. Due to their simplicity, cost-effectiveness, and reliability, the developed spectrophotometric methods are suitable for routine quality control analysis of Meropenem in pharmaceutical industries.

Keywords: Meropenem, Visible spectrophotometry, Chromogenic reagents, Method development, Method validation, Beer–Lambert's law, Pharmaceutical analysis, Ion-association complex, Oxidative coupling, RP-HPLC

Introduction

Drugs are defined as substances used for diagnosis, prevention, and treatment of diseases, or for modifying physiological functions in the body [1–5]. The development of modern pharmaceutical science has led to the discovery and formulation of a wide range of synthetic and biologically derived therapeutic agents. The transformation of a chemical compound into a pharmaceutical product involves three essential stages: drug discovery, bulk manufacturing, and formulation into suitable dosage forms such as tablets, capsules, and injections.

Pharmaceutical analysis plays a crucial role in ensuring the quality, safety, and efficacy of drug substances and their formulations. It involves qualitative and quantitative evaluation of drugs, impurities, and excipients using various analytical techniques Regulatory authorities such as pharmacopoeial bodies establish strict guidelines to maintain drug quality standards [6–8]. Among various analytical approaches, physicochemical methods such as spectrophotometry and chromatography are widely used due to their accuracy, sensitivity, and rapid analysis[6-8].

Spectrophotometry is one of the most extensively employed analytical techniques in pharmaceutical analysis. It is based on the measurement of absorption of electromagnetic radiation by molecules, which is directly related to their concentration. The UV-visible spectrophotometric method is particularly advantageous due to its simplicity, cost-effectiveness, and applicability for both colored and colorless substances. The principle of spectrophotometric analysis is governed by Beer–Lambert's law, which states that absorbance is directly proportional to the concentration of the absorbing species and the path length of the medium:

Where A is absorbance, ε is molar absorptivity, b is path length, and c is concentration. In pharmaceutical analysis, visible spectrophotometric methods often involve the formation of colored complexes through chemical reactions such as oxidation-reduction, charge transfer, ion-association, condensation, and oxidative coupling reactions. These reactions enhance sensitivity and selectivity, allowing the determination of drugs even at low concentrations. Chromogenic reagents such as DDQ, p-chloranilic acid, MBTH, and various dyes are widely used for such purposes literature reveals that several UV spectrophotometric methods have been developed for the estimation of different drugs in pharmaceutical formulations, showing good accuracy and precision within Beer's law limits However, visible spectrophotometric methods offer distinct advantages by minimizing interference from excipients that absorb in the UV region.

Meropenem is a broad-spectrum β-lactam antibiotic widely used in the treatment of severe bacterial infections. Due to its clinical

importance, accurate and reliable analytical methods are essential for its quality control. Although several analytical methods have been reported, there is still a need for simple, economical, and sensitive methods suitable for routine analysis in pharmaceutical laboratories. Therefore, the present study aims to develop and validate new visible spectrophotometric methods based on different chromogenic reactions for the determination of Meropenem in bulk and pharmaceutical dosage forms. These methods are designed to be simple, rapid, cost-effective, and suitable for routine quality control applications.

Materials and Methods

Instruments used:

Genesys 10 UV-Spectrophotometer 10mm matched quartz cells procured from Thermo Scientific company with was used for all spectral measurements. A Systronics digital pH meter [Model-362] was used for pH measurements.

Preparation of reagents:

All the chemicals and reagents used were of analytical grade and solutions were prepared in doubled distilled water.

DDQ solution (Loba 0.2% w/v):

Prepared freshly by dissolving 200mg of DDQ in 100mL of acetonitrile (Merck, Mumbai, India).

PCA solution (Loba 0.1% w/v)]:

Prepared by dissolving 100mg of para chloranilic acid in 100mL of 1,4-dioxane (Merck, Mumbai, India)was prepared and kept in the dark when not in use.

4-AP reagent (BDH; 0.5%)]:

Prepared by dissolving 500mg of 4-AP in 100mL of MeOH containing 1.0% of conc HCl.

INH reagent (Sd-Fine; 0.8%)]:

Prepared by dissolving 800mg of INH in 100mL of MeOH containing 1.0% of conc HCl.

Anthranilic acid, (Loba; 0.25%)]:

Prepared by dissolving 0.25g of anthranilic acid in 100mL methanol.

2-chlorophenyl hydrazine, (Loba; 0.25%) reagent]:

Prepared by dissolving 0.25g of 2-chlorophenyl hydrazine in 100mL methanol.

0.1%,2,4-Dinitro Phenyl Hydrazine Reagent: Prepared freshly by dissolving 100mg of 2,4-Dinitro Phenyl Hydrazine in a mixture of 10mL of methanol and 0.5mL of Conc.HCl was added and finally diluted to 100mL with methanol.

Bromo Thymol Blue Solution:

A 0.1% solution of Bromothymol blue (BTB) was prepared by dissolving the 100mg dye (Rankem, India, 95% dye content) in water and filtered to remove the insoluble residue.

Bromocresol Purple solution (0.2% W/V):

Prepared by dissolving 200mg of BCP dye in 100ml of double distilled water.

Phthalate buffer (pH 3.5) was prepared by adjusting the pH of 0.1 mol.L–1potassium hydrogen phthalate with 0.1mol.L–1 hydrochloric acid and 0.1mol.L–1 sodium hydroxide, respectively.

MBTH solution (Loba, 0.5%, 2.14 x 10-2M):

Prepared by dissolving 500mg of MBTH in 100mL distilled water.

NaOH Solution (Loba, 0.4%, 0.1M):

Prepared by dissolving 400mg of NaOH to 100mL distilled water and standardized.

Fe (III) Solution (Merck, 0.25%, 1.53x10-2M): Prepared by dissolving 250mg of Ferric chloride in 100mL distilled water.

HCl Solution (Qualigens, 1.0M):

Repared by dissolving 8.6mL of conc. HCl to 1000mL distilled water and standardized.

Preparation of Standard Drug Solution:

0.1% stock solution of Meropenem was freshly prepared by transferring accurately weighed 100mg of Meropenem into 100mL volumetric flask and dissolved in double distilled water, and then made up to the mark. Then working standard solutions were prepared by transferring aliquots of the stock solution into a series different 100mL volumetric flasks respectively and made up to the mark was used for the following methods.

Procedure for dosage forms:

500mg of meropenem powder procured from local pharmacy was dissolved in about 10.0mL of ethanol and filtered through a Whatman No.42 filter paper. The filtrate was made up to mark with distilled water in a 100mL volumetric flask. Suitable volumes of this filtrate was pipetted and diluted up to the mark with distilled water. Aliquots of these solutions were used for the determination of Meropenem drug as per the procedures described.

Proposed procedures:

DDQ: Aliquots (0.5 -3.0mL) of a standard meropenem (80µg.mL-1) solution were accurately transferred into a series of 10.0mL volumetric flasks and the total volume was adjusted to 3.0mL by adding adequate quantity of acetonitrile to each flask. Then 2.0mL of 0.2% DDQ solution was added to each flask and the mixture was diluted to the volume with acetonitrile and the absorbance of each solution was measured at 560nm against a reagent blank.

PCA: Different aliquots of standard meropenem solutions (0.5 - 2.5mL;50µg.mL-1) were accurately transferred into a series of 10.0mL volumetric flasks and the total volume was adjusted to 3.0mL by adding adequate quantity of acetonitrile. To each flask 2.0mL of 0.1% p-chloranilic acid was added, and the contents were mixed well and kept aside for 10min. The mixture was diluted to the volume with acetonitrile and the absorbance was measured at 520nm against a reagent blank prepared simultaneously. The concentration of the meropenem was read from the standard graph using the Beer‘s law data.

4-AP:

Deliver aliquots of standard meopenem solution (0.5- 2.5mL, 40mg.mL-1) were transferred into a series of 10mL calibrated tubes. Then 3.0mL of 4-AP was added to each tube and kept aside for 15min. Later the solution in each tube was made up to 10mL with methanol. The absorbance was measured at 450nm against the reagent blank. The amount of meropenem was computed from its calibration graph.

INH:

Deliver aliquots of standard meopenem solution (0.5 - 2.5mL, 80 g.mL-1) into a series of 10mL calibrated tubes. Then 2.0mL of INH solution was added to each tube and heated for 10min at 60oC. Later the solution in each tube was made up to 10mL with methanol. The absorbance was measured at 465nm against the reagent blank. The amount of meopenemr was computed from its calibration graph.

ATA & 2-CPH:

Aliquots of (0.5-2.5mL, 40 g.mL-1) drug were transferred into a series of 10.0mL volumetric flasks. To each of the above

aliquots, 2.0mL of anthranilic acid was added followed by one drop of concentrated hydrochloric acid and heated to 50-55oC for color development. The same procedure was carried out for 2-Chloro phenyl hydrazine (0.5- 2.5mL, 40 g.mL-1). The absorbance's of the color derivatives were measured at 510nm for anthranilic acid, 550nm for 2-chlorophenyl hydrazine against reagent blank. The amount of drug was computed from its calibration graph (Fig. 4.16 & 4.17).

2,4-DNPH:

Aliquots of working standard meopenem solution ranging from 0.5- 2.5mL (40 g.mL-1) were transferred into a series of 10.0mL calibrated test tubes. To this 2.0mL of 2,4-DNPH reagent was added followed by one drop of Conc.HCl. The mixtures were placed on a boiling water bath for 10min and were cooled to room temperature. The contents of the tubes were mixed thoroughly and allowed to stand for 10minutes with occasional shaking at room temperature and the final volume in each tube was made up to the mark with distilled water. The pink colored chromogen was measured spectrophotometrically at 480nm against a reagent blank.

BTP:

Aliquots of working standard solutions of meopenem (0.5-3.0mL,80mg.mL-1) were taken in different 125mL separating funnels. To each of the separating funnels BTB dye solution (5.0mL), buffer solution (5.0mL,pH-3.5)and chloroform (10.0mL) were added and the contents were mixed for 2min. The layers were allowed to separate. The separated layers were collected in dry test tubes and the absorbance of each organic layer was measured in 1.0cm cell at 420nm against blank. The concentration of the unknown was read from the calibration graph or computed from the regression equation.

BCP:

Aliquots of working standard solutions of meopenem (0.5-3.0mL, 80 g.mL-1) were taken in different 125mL separating funnels. To each of the separating funnels BCP dye solution (5.0mL), buffer solution (5.0mL, pH-3.5) and chloroform (10.0mL) were added and the contents were mixed for 2min. The layers were allowed to separate. The separated layers were collected in dry test tubes and the absorbance of each organic layer was measured in 1.0cm cell at 418nm against blank. The concentration of the unknown was read from the calibration graph from the regression equation.

MBTH-Fe (III):

Standard solutions of meropenem having concentrations in the range (0.5-3.0mL, 100 g.mL-1) were transferred into a series of 10.0mL volumetric flasks. To each 2.0mL of MBTH, 2.0mL of ferric chloride was added and the volume was made up to mark with distilled water and allowed to stand for 20minutes. The absorbance of each solution was measured at 600nm against the reagent blank. The colored species was stable for 2hours and the amount of drug in the sample was computed from its calibration curve represented.

Results and Discussion

Parameters Fixation (Optimization Studies):

The Optimization studies for the proposed procedures involve the study of the influence of various factors on the color development [optimal conditions] such as reagent concentration, order of addition of reagents, time, temperature and choice of solvent for maximum color development.

4-AP:

The optimum conditions in this method were established, based on the study of the effects of various parameters such as volume of 4-AP solution, volume of solvents solution used initially and subsequently for final dilution and the stability of colored species after final dilution. The optimum conditions developed and actual conditions chosen for the procedure are recorded.

INH: The optimum conditions for this method was made based on the study of the effects of various parameters such as volume of INH solution, volume of solvents solution used initially and subsequently for final dilution and the stability of colored species after final dilution. The optimum conditions developed and actual conditions chosen for the proposed method are recorded.

ATA & 2-CPH: The optimum conditions for the proposed methods were found basing on the study of the effects of various parameters such as volume of INH solution, volume of solvents solution used initially and subsequently for final dilution. The optimum conditions developed and actual conditions chosen for the procedure are recorded.

2,4-DNPH:

The optimum conditions in this method were established basing on the study of the effects of various parameters such as volume of 2,4-DNPH solution, volume of solvents solution used initially and subsequently for final dilution and the stability of colored species after final dilution. The optimum conditions developed for the proposed method is recorded.

BCP:

This method involves the formation of ion-association complex between meropenem and BCP. In order to establish the optimum conditions necessary for rapid and quantitative formation of the colored product with maximum stability and sensitivity, the author performed controlled experiments by measuring absorbance at 420nm of a series of solutions, varying one and fixing the other parameters in each case such as type and volume of acid, concentration of dye, organic solvent used for extraction, ratio of organic phase to aqueous phase during extraction, shaking time and temperature. The results are incorporated.

MBTH-Fe(III):

The method involves the reaction of meropenem with MBTH in the presence of an oxidant, Fe (III) solution. The optimum conditions in this method were fixed, basing on the study of the effects of various parameters such as MBTH and alkali to be added, acid and oxidant required after cooling and the solvent used for final dilution on the intensity and stability of the colored species formed. The optimum conditions found and actual conditions chosen for the procedure are incorporated.

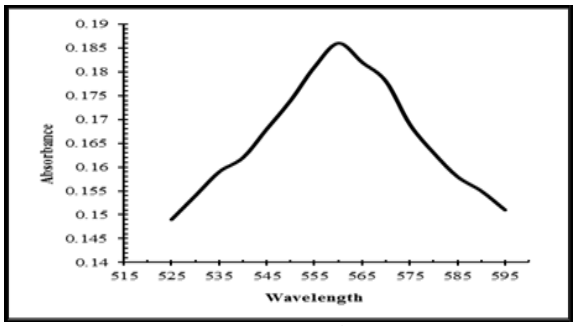

Fig.1: Absorption spectrum of MPM with DDQ

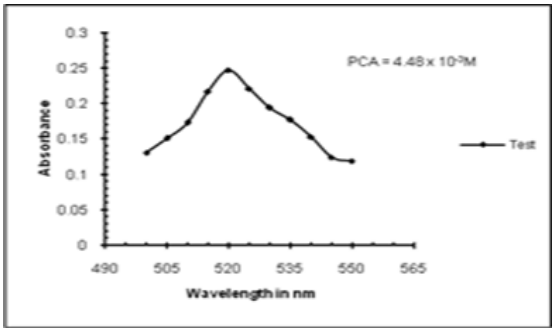

Fig. 2: Absorption spectrum of MPM with 4-AP

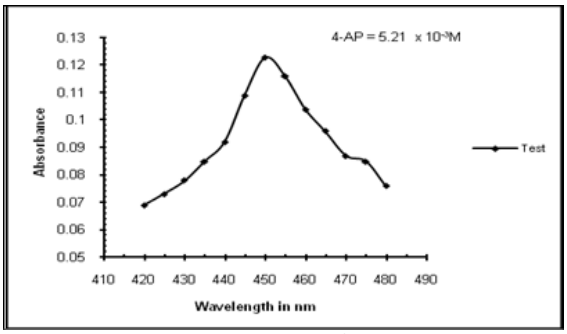

Fig. 3: Absorption spectrum of MPM with INH

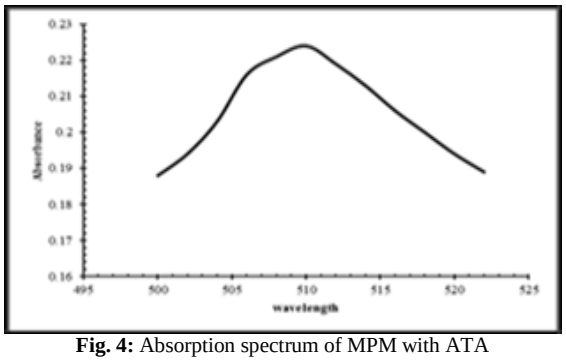

Fig. 4: Absorption spectrum of MPM with ATA

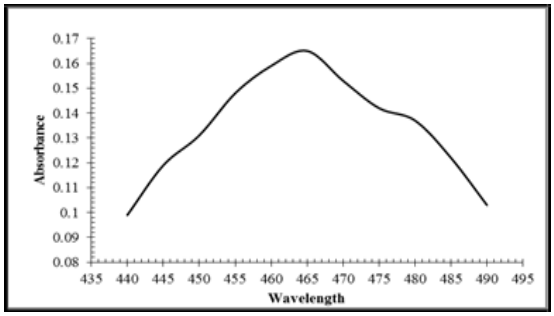

Fig. 5: Absorption spectrum of MPM with 2-CPH

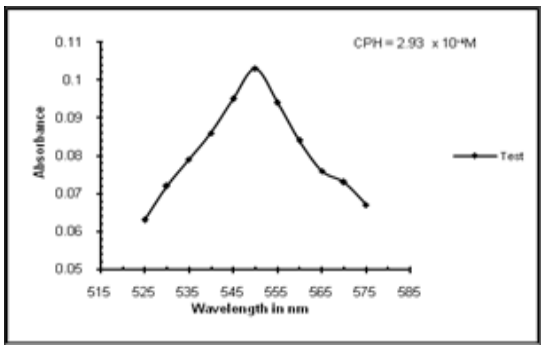

Fig.6: Absorption spectrum of MPM with 2,4-DNPH

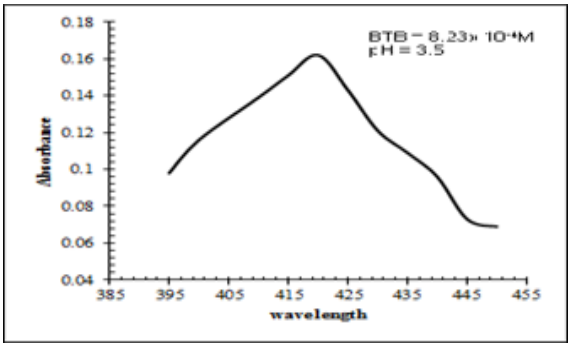

Fig.7: Absorption spectrum of MPM with BTB

Discussion

In the present investigation, several visible spectrophotometric methods were developed for the quantitative determination of Meropenem using different chromogenic reagents. Each reagent formed a stable colored complex with the drug under optimized conditions, allowing accurate spectrophotometric measurement. The spectral characteristics of the developed methods showed distinct absorption maxima at different wavelengths such as 560 nm for DDQ, 520 nm for PCA, 450 nm for 4-AP, 465 nm for INH, 510 nm for anthranilic acid, 550 nm for 2-chlorophenyl hydrazine, 480 nm for DNPH, 420 nm for BTB, 418 nm for BCP, and 600 nm for MBTH-Fe (III). These wavelengths correspond to the formation of colored chromogenic complexes between Meropenem and the reagents. Beer's law was obeyed over different concentration ranges depending on the reagent used. The regression coefficients for all methods were found to be close to 0.999, indicating excellent linearity and reliability of the developed analytical methods.

The calculated values of molar absorptivity and Sandell's sensitivity demonstrated good sensitivity of the proposed methods. Precision studies revealed low relative standard deviation values, confirming the reproducibility of the methods. Accuracy studies performed using recovery experiments showed percentage recoveries close to 98–100%, indicating that the methods are highly accurate. The developed methods were successfully applied to the analysis of commercial pharmaceutical formulations containing Meropenem. The assay results obtained by the proposed methods were in good agreement with those obtained by the reference method. Statistical comparison using t-test and F-test indicated no significant difference between the proposed and reference methods at the 95% confidence level. Thus, the proposed visible spectrophotometric methods proved to be reliable for the routine analysis of Meropenem in pharmaceutical preparations.

Conclusion

The present investigation successfully developed several visible spectrophotometric methods for the quantitative determination of Meropenem in bulk drug and pharmaceutical dosage forms. The proposed methods are simple, sensitive, accurate, and reproducible. They require inexpensive reagents and simple instrumentation, making them suitable for routine quality control analysis. Unlike many reported UV spectrophotometric methods, the developed methods involve measurements in the visible region, which minimizes interference from UV-

absorbing excipients present in pharmaceutical formulations. The results obtained from the analysis of pharmaceutical formulations showed good agreement with the reference method, confirming the reliability of the proposed procedures. Therefore, the developed visible spectrophotometric methods can be effectively used as alternative analytical methods for the routine determination of Meropenem in bulk and pharmaceutical dosage forms in quality control laboratories.

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Conflict of Interests
The authors declare no conflict of interest

Ethics Approval Not applicable

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AI Tool Declaration
The authors declare that no AI and related tools are used to write the scientific content of this manuscript.

Data Availability
Data will be available on request

References
[1] Satoskar RS, Bandarkar SD, Rege NN, Ainapure SS. Pharmacology and Pharmacotherapeutics. 15th ed. Mumbai: Popular Prakashan, 1996.
[2] Wolff ME. Burger's Medicinal Chemistry. 4th ed. New York: Wiley Interscience, 1981.
[3] Doerge RF. Wilson and Gisvold's Textbook of Organic Medicinal and Pharmaceutical Chemistry. 8th ed. Philadelphia: Lippincott Company, 1982.
[4] Angrejus K. Essentials of Medicinal Chemistry. 2nd ed. New York: Wiley Interscience, 1988.
[5] Goodman LS, Gilman A, Rall TW, Nies AS, Taylor P. The Pharmacological Basis of Therapeutics. 8th ed. New York: Pergamon Press, 1990.
[6] Topliss JG. Quantitative Structure–Activity Relationships of Drugs. London: Academic Press, 1983.
[7] Foye WO. Principles of Medicinal Chemistry. 3rd ed. Bombay: Lea & Febiger, 1989.
[8] Government of India. Drugs and Cosmetics Act and Rules. New Delhi: Government of India Publications, 1984.
[9] Indian Pharmacopoeia Commission. Indian Pharmacopoeia. New Delhi: Ministry of Health and Family Welfare, 1996.
[10] British Pharmacopoeia Commission. British Pharmacopoeia. London: HMSO, 2002.
[11] Reynolds JEF. Martindale: The Extra Pharmacopoeia. 33rd ed. London: Pharmaceutical Press, 2002.
[12] Chatten LG. Pharmaceutical Chemistry. New York: Marcel Dekker, 1996.
[13] Beckett AH, Stenlake JB. Practical Pharmaceutical Chemistry. 4th ed. New Delhi: CBS Publishers, 1986.
[14] Perez J, Bartol J. Colorimetric and Fluorimetric Analysis of Organic Compounds and Drugs. New York: Marcel Dekker, 1974.
[15] Sastry CSP, Lingeswara Rao JSVM. Spectrophotometric determination of pharmaceutical compounds. Talanta. 1996, 43: 1827–1835.
[16] Snell FD, Snell PT. Colorimetric Methods of Analysis. London: Van Nostrand, 1959.
[17] Mohamed GG, Nour El-Dien FAF, Mohamed NA. Spectrophotometric determination of drugs. Spectrochim Acta A. 2007, 67: 1244–1249.
[18] Darwish IA. Analytical spectrophotometric methods. Anal Chim Acta. 2005, 549: 212–220.
[19] Sivasubramanian VL, Kasi Sankar V, Sivaraman V, Senthil Kumar K, Muthukumaran T, Raja TK. Spectrophotometric estimation of pharmaceutical compounds. Indian J Pharm Sci. 2004;66(6):799–802.
[20] Mohamed GG, Nour El-Dien FAF, Mohamed NA. Spectrophotometric determination of drugs. Spectrochim Acta A Mol Biomol Spectrosc. 2007; 67: 1244–1249.
[21] Hasani M, Akbari S. Spectrophotometric determination of pharmaceutical compounds. Spectrochim Acta A Mol Biomol Spectrosc. 2007;68(3):409–413.
[22] Wu H, Du LM. Spectrophotometric determination using chromogenic reagents. Spectrochim Acta A Mol Biomol Spectrosc. 2007;67(3–4):976–979.
[23] Walash MI, Sharaf-El Din M, Metwalli MES, Shabana MR. Spectrophotometric determination methods. Arch Pharm Res. 2004; 27: 720–726.
[24] Khashaba PY, El-Shabouri SR, Emara KM, Mohamed AM. Spectrophotometric determination of pharmaceutical compounds. J Pharm Biomed Anal. 2000; 22:363–376.
[25] Abdellatef HE. Spectrophotometric analysis of pharmaceutical drugs. J Pharm Biomed Anal. 1998; 17: 1267–1271.